REPORT OF EXPERIMENTS NOW BEING PURSUED IN THE BACTERIOLOGICAL LABORATORY OF THE ACADEMY.

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Experiments show that the twenty-fifth and the second growths of the tubercle bacillus will multiply with great rapidity when planted on a nutrient medium made up of:—

Extract of Beef	25.g
Aq: Dist:	700.cc
Dry peptone	8.g
Agar-agar	15.g
Chlor. Soda	5.g
Dist. Water (added.)	300.cc
Glycerine	100.00

The turbidity is removed by the slow mixing of the whites of two hens eggs.

If four distinct needle tracks are made with these bacilli lengthwise in a half-inch tube containing this medium, the entire surface will be covered with a thin layer of tuberculous growth within five weeks. The bacilli from the twenty-fifth culture present a variety of forms, some club-shaped, others branched, while many would appear to grow to a great length, certainly measuring in some instances three times that of the average tubercle bacillus described by the authorities on this organism.

To obtain the toxic substance of bacilli, they may be floated in a liquid medium. By this method we can filter out the toxic substance without submitting the medium to heat. With some of the toxic agents generated by aerobic micro-organisms, this method will prove of vital importance, as heat destroys many of the ptomaines.

The tuberculous poison is most readily and cheaply obtained from tuberculous lungs of cows and the viscera of other animals. Before giving a resume of the simple method of obtaining the toxic agent from the lungs, I wish to thank Dr. Francis Bridge, State Veterinary Inspector, for his kindness in furnishing the Laboratory with tuberculous material. It is owing to his courteous attention that I have never wanted for tuberculous lungs of cows since 1889. He has supplied the numerous demands at the shortest possible notice, and has insisted upon contributing his time and trouble for the benefit of science.

The tuberculous mass is first placed in a mortar and then finely chopped up with a pair of shears. Then it is thoroughly mashed with a pestle, and treated with water, or better still with water and glycerine, for twelve hours at a low temperature. It has been my habit, however, to expose it to a temperature of 40° C. for twelve hours longer; after which the substance is poured into a Chamberlain-Pasteur filtering tube, which it is permitted to slowly pass through without pressure.

Its effect on tuberculous animals would seem to be about as represented by the following reaction:—

Tuberculous Cow. Before inoculation temperature 101° F. 3 cc. of filtrate was subcutaneously injected at 10 a. m. At 5 p. m. temperature was 102°; at 10 p. m. 103°; at 12 p. m. 103⅘°; at 10 a. m. 101⅗°.

Guinea Pigs.[1] No. 3-A. At 11 a. m. the temperature was 101⅗°; at 1 p. m. 101°; at 3 p. m. 101°. At 2.20 p. m. 1–20 cc. of tuberculous agent from lung was subcutaneously injected. At 5.45 the temperature was 101⅕°; at 7 p. m. 102°; at 11 p. m. 101⅘°; at 1.30 a. m. 101⅗°; at 8 a. m. 101 ⅗°; at 12.30 p. m. 100⅘°; at 3 p. m. 101⅘°.

No. 4-A. At 11 a. m. 101⅕°; at 1 p. m. 101⅘°; at 3 p. m. 101⅗°: At this time 1–10 cc. toxic agent from lung of a cow subcutaneously injected.

At 5.45 p. m. temperature 102⅘°; at 7 p. m. 103⅕°; at 11 p. m. 102⅖°; at 1.30 p. m. 101⅘°; at 8 a. m. 101⅗°; at 12.30 p. m. 102⅕°; at 3 p. m. 101⅘°.

In addition to my last report of investigations I have to refer to the treatment of tuberculosis by subcutaneous injections of dilute sulphuric acid and formic acid. Under each of these methods the animals would appear to be doing better than the check guinea pigs not so treated, yet sufficient time has not elapsed for me to make any post-mortems to determine the processes going on in the viscera. The points of inoculation, however, have healed beautifully and the animals would appear to be doing well. The experiments towards the securing of immunity are still being conducted and the results certainly show that the animal organism can be cultivated to resist inoculations of the tubercle bacillus to a greater or less

[1] Temperature in the Guinea pig is not constant; and therefore not as reliable for experimentation as either the cow or dog.

degree, as is shown by a copy of the record book in a few of the cases under treatment.

Rabbit D–100. Jan. 10th, 1891. Inoculated with a mass of twenty-fifth artificial cultivation of bovine tubercle bacillus.

Feb. 16th, 1891. Inoculated again from the same tube as in January.

Feb. 28th, 1891. Inoculated with a mass of second artificial cultivation of bovine tubercle bacilli.

At this time the animal presents a healthy appearance and the points of inoculation have healed up.

Rabbit D–101. Showed the same result as rabbit D–100

Without going into further detail, I will simply state that a number of guinea pigs have given like results; whereas the animals inoculated with a mass of the second cultivation of bovine tubercle bacilli, not having been previously inoculated with the attenuated virus, sickened with tuberculosis and in some instances died.

The last experiments confirm the fact that the tubercle bacillus does lose its virulence to a certain degree by cultivating it continuously on an artificial medium.

Dog C–1. Eight weeks old, weight 1½ lbs. Was inoculated with second artificial culture of bovine tubercle bacillus and died within ten days, of toxæmia.

Dog C–2. Same age and weight as C–1. Under the same treatment, also sickened with tuberculosis.

Dog C–3. Same age and weight as C–1. Was at the same time inoculated with twenty-fifth artificial culture of bovine tubercle bacillus. While the others sickened with tuberculosis, this one shows no outward manifestation of the malady.

In one of my recent communications, I referred to the fact that I had been injecting predigested fat plus antiseptics into the intestinal tract of animals suffering from tuberculosis with some success and also that I was subcutaneously introducing some of the bile salts. The results from the bile salts have not been at all constant; while the subcutaneous injection of glycerine into the tuberculous animals, which I recently referred to in a Medical Journal of this city, has apparently given a result which will stimulate work in this direction.

Two dogs eight weeks old had respectively a mass of the second cultivation of bovine tubercle bacilli subcutaneously introduced. In seven days one was dead apparently from the toxic effect of the product of the bacilli.

The day that this dog died, the other presented a line of symptoms indicating toxæmia to a very marked degree. The animal was hardly able to stand and I had every reason to believe it was fast losing its vitality.

One-half cc. of pure glycerine, well diluted, was subcutaneously introduced. In a short time the animal showed physical improvement and in twelve hours it was restored to apparently a good condition. It would seem that the glycerine enabled the viscera to get rid of the toxic agent that otherwise would have passed into the general circulation.